10-83

Attallah Kappas, M.D.

METABOLISM-PHARMACOLOGY

Dr. Attallah Kappas, Sherman Fairchild Professor of The Rockefeller University, is physician-in-chief of The Rockefeller University Hospital and head of the Metabolism-Pharmacology Laboratory. He is a leading authority on metabolic liver diseases, biochemical and clinical pharmacology, and environmental medicine. At present, Dr. Kappas is involved in research that he hopes will result in a new and better treatment for postnatal jaundice, a prevalent and sometimes lethal condition in newborn infants.

Heme and Heme Oxygenase

The liver, one of the largest organs in the human body, serves as the primary receiving depot, chemical processing plant, and distribution center for drugs and foreign chemical substances. One important component of the liver's purification system is the iron compound heme--the red pigment of blood.

Heme combines with a protein (globin) to make hemoglobin, which transports oxygen in the blood throughout the body. It also combines with proteins in liver cells to produce cytochromes, which play a vital role in the body's defense against chemical agents. These agents may be products of the body, such as steroid hormones, or they may enter the body from the environment in the form of drugs, food components, and environmental pollutants. Without the regulatory processes controlled by cytochromes, particularly a specific cytochrome called P-450, many chemicals might continue to act indefinitely on the body's organs, with harmful consequences.

When a red blood cell or cytochrome is broken down, the heme molecule is opened, and the iron atom bound in the center of its molecular structure is released. Known as heme degradation, this activity involves a specific enzyme--a protein that accelerates the catalytic action. This enzyme, heme oxygenase, controls the rate at which heme molecules are degraded.

Heme Oxygenase and Neonatal Jaundice

When heme degradation takes place, the iron molecule is usually salvaged by the liver for future use, while the rest of the molecule is converted to bile pigments and then excreted. The immediate product of heme degradation is a green pigment, biliverdin. In some vertebrates, this water soluble and nontoxic compound is simply excreted by the liver in bile and urine.

Mammals transform biliverdin to a fat-soluble substance, bilirubin, prior to excretion. It is believed that this transformation evolved as part of the mammal's reproductive system. Because bilirubin is fat soluble, it is able to cross the fatty membranes lining the cells of the placenta, which connects the fetus to the uterus of its mother. The advantage of this arrangement is that bilirubin produced by the fetus can pass from the fetus into the mother's circulatory system, where it is then processed and excreted.

Humans infants, however, cannot rid themselves of bilirubin during the first two weeks of life, because their livers are deficient in the enzyme that can conjugate bilirubin and permit its excretion into bile. This leads to the accumulation of bilirubin in the plasma shortly after birth, producing neonatal jaundice, or hyperbilirubinemia. In severe form, this condition can damage various types of cells, especially brain cells. Until the liver matures, high levels of bilirubin in the blood remain potentially dangerous for the child, since the blood/brain barrier, which excludes many chemicals from the central nervous system, is also immature and permeable to toxic substances such as bilirubin.

In neonatal jaundice the effects of bilirubin on the central nervous system comprise a syndrome referred to as "kernicterus." The clinical manifestations of this syndrome include lethargy, weakness of skeletal muscles, poor feeding due to loss of the sucking reflex, convex bending of the spine, spasticity, and death. The brains of infants who die from postnatal hyperbilirubinemia show yellow pigmentation (i.e., bilirubin deposition) in key areas of the brain. Survivors of the disorder show a high incidence of nerve deafness and abnormal mental development, and long-term studies reveal subtle neurological and psychological abnormalities even in apparently normal infants who accumulate high levels of bilirubin in their blood after birth.

The importance of these findings cannot be underestimated since they suggest strongly that bilirubin toxicity may well be a "no-threshold" phenomenon. That is, during the first four weeks after birth, <u>any</u> level of bilirubin in the blood higher than normal may be potentially harmful, even though some amount of the substance is inevitable.

Since brain cells do not regenerate, it is impossible to determine the actual influence of bilirubin, even at relatively low levels, on neurological development. It seems likely, however, that the higher the level of bilirubin, the greater the chance of damage to the brain and the central nervous system.

Laboratory Research

Dr. Kappas and his colleagues are studying the relationship between chemical reactions involving heme oxygenase and the formation of bilirubin. In contrast to current therapies for neonatal jaundice and other hyperbilirubinemic disorders, which attempt to dispose of bilirubin after it is formed, scientists at The Rockefeller are working to block bilirubin production before it starts.

In one investigation, heme oxygenase was successfully isolated and purified by Dr. Kappas, Dr. Takeo Yoshinga, a visiting scientist from Japan, and Dr. Shigeru Sassa, an Associate Professor in Dr. Kappas's laboratory. Studies of purified heme oxygenase and the mechanics of heme oxidation have demonstrated that certain trace metal compounds affect the metabolic sequence of heme degradation without altering heme oxygenase. These studies have led to the belief that a simple, effective, and safe chemical might be found to regulate heme degradation, inhibit bilirubin formation, and thus prevent postnatal jaundice and other hyperbilirubinemic disorders.

This theory was confirmed in related investigations by Dr. Kappas and Dr. George S. Drummond, an Assistant Professor in the Kappas laboratory. Their research has shown that synthetic heme containing tin, rather than iron, blocks the binding of heme to heme oxygenase and thus prevents the formation and subsequent accumulation of bilirubin in the liver, spleen, and other tissues of experimental animals.

Tin-heme is similar to iron-heme, differing only in the substitution of a tin atom for an iron atom in the molecule. Experimental research indicates that tin-heme and heme oxygenase will combine at a rate of at least 100 times greater than iron-heme and heme oxygenase. As a result, tin-heme blocks iron-heme's access to the enzyme. Tin-heme cannot be converted to bilirubin by heme oxygenase, and the entire process of iron-heme conversion to bilirubin is prevented until tin-heme is slowly excreted from the body. Tin-heme is not toxic. It does not interfere with other iron-heme functions such as the transport of oxygen. It does not enter the brain, and it appears to be released from the liver and spleen unaltered.

Neonatal hyperbilirubinemia in rats has been shown to be completely prevented by small doses of tin-heme administered to newborn rats immediately after birth. No toxic side effects have been observed in the newborn rats or in adult animals--even with doses 100 times greater than those that have proved effective in blocking postnatal jaundice. In addition, the offspring of rats who have been treated with tin-heme show no side effects from the drug.

Tin-heme may have significant potential for clinical use in preventing the formation of bilirubin in newborn babies who might otherwise suffer neurological impairment, irreversible brain damage, or death. Tin-heme therapy may also prove to be effective in regulating bilirubin production in other clinical circumstances in which this may be desirable.

Clinical studies of adult patients have been approved by the Federal Drug Administration and are currently in progress at The Rockefeller University Hospital. Initial studies will be limited to adults. Successful results may then make it possible to extend treatment to infants with neonatal jaundice, who will benefit most from the therapy.

October 1983